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ORIGINAL ARTICLE

Density, viscosity, and refractive index of mono-, di-, and tri-saccharides in aqueous glycine solutions at different temperatures

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Abstract Density, ρ, viscosity, η, and refractive index, n_D of glucose, sucrose, and raffinose have been measured in 0.015 *m* aqueous glycine at 298.15, 303.15, 308.15, and 313.15 K. From these experimental data, apparent molar volumes, V_ϕ, apparent specific volumes, $V_{\phi,sp}$ as a function of the concentration of solutes, the standard partial molar volume, V_ϕ^0, transfer volume from water to aqueous glycine solutions, $\Delta V^0_{\phi(tr)}$, and partial molar expansibility of solute, E_ϕ^0 have been calculated for glucose, sucrose and raffinose in aqueous glycine solutions. Falkenhagen coefficient A and Jones–Dole coefficient B, free energies of activation of viscous flow per mole of solvent, $\Delta\mu_1^{0\#}$, and solute, $\Delta\mu_2^{0\#}$, enthalpy, $\Delta H^{0\#}$, and entropy, $\Delta S^{0\#}$ of activation of viscous flow have been evaluated by using viscosity data. The molar refraction has been calculated by using measured refractive index data. Results have been explained in terms of solute–solute and solute–solvent interactions.

1. Introduction

Saccharides are the most abundant and diverse classes of organic molecules found in nature and their conjugates with proteins and lipids play key roles in the immune and endocrine systems, fertilisation, brain development, prevention of pathogenesis and blood clotting (Safina, 2012). They serve as energy sources (Safina, 2012; Wang et al., 2012; Knapp et al., 2008) and play a very crucial role in biological recognition phenomena in the process of exchanging information between the cells like cell–cell interactions and cell death signal transduction inflammatory processes, cancer metastasis bacterial and viral infections, fascinating the researchers to design a new category of anticancer drugs (Safina, 2012; Carroll et al., 2006; Kaminski et al., 2012). Recently, Montesarchio and co-workers designed saccharide based synthetic-ion transporters which are of great interest for both technological and biomedical applications (Montesarchio et al., 2012). Interactions of saccharides with proteins play an important role in a wide range of biochemical processes, for example, the application of saccharides as lyoprotectant. It is well-known that there are a lot of protein pharmaceuticals which are prepared by

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freeze-drying. However, in many cases this process breaks the native structure of protein and leads to its destabilisation. Addition of saccharide minimises the unwanted processes of freeze drying. Due to the great ability of saccharides to form hydrogen bonds with target molecules as well as their weak tendency to recrystallize, aggregation and destabilisation of proteins can be completely avoided (Kaminski et al., 2012). Understanding the mechanism of protein–saccharide interaction and the subsequent stabilisation of proteins by saccharide molecules is still incomplete (Karlsson, 1987; Osawa and Tsuji, 1987). However, due to the complex conformation and configuration of proteins in various solvents, a direct study on proteins is difficult, so we select glycine, the simplest amino acid and constituent of proteins to study the thermodynamic properties of saccharide–amino acid interactions in aqueous solutions. Literature survey indicates that interactions between saccharides and proteins have been investigated using chromatography data, (Karlsson, 1987; Osawa and Tsuji, 1987) X-ray crystallography,(Quiocho and Vyas, 1984; Bundle, 1989) NMR spectra, computer simulations, (Thøgersen et al., 1982; Bock, 1983) and kinetics (Miller et al., 1983; Miller et al., 1980). However, thermodynamic and transport studies of interactions of saccharides with model compounds of proteins (amino acids) are rare. Interactions between glucose and amino acids (glycine, alanine, serine, and valine) (Ali et al., 2006) using volumetric, viscometric, and refractive index techniques, and those between disaccharides (D-maltose and sucrose) and amino acids (glycine, alanine, leucine, and serine) (Parfenyuk et al., 2004) using calorimetric titrations have been reported in dilute aqueous solutions. Such studies can provide better and valuable information towards understanding the behaviour of these biomolecules in aqueous media. Lack of thermodynamic and transport studies on saccharides in aqueous amino acids led us to investigate saccharide–amino acid interactions through the determination of various volumetric and viscometric parameters of glucose, sucrose, and raffinose in aqueous glycine at different temperatures. The derived parameters are used to discuss saccharide–glycine and saccharide/glycine–water interactions.

2. Materials and methods

High purity glycine (E. Merck, Germany, mass fraction 0.99) was used after recrystallisation from ethanol + water mixture and dried over P_2O_5 in a vacuum desiccator. D-glucose (Thomas Baker Chem. Ltd., Mumbai, India, mass fraction 0.99), sucrose (E. Merck, Germany, mass fraction 0.99) and D- (+)- raffinose pentahydrate (raffinose) (Sigma Aldrich, USA, mass fraction 0.99) were used as such, except drying them under vacuum at 40°C for two days. Aqueous solution of 0.015 m (mol kg^{-1}) of glycine was prepared using deionised double distilled degassed water (conductivity 1.3×10^{-6} S cm^{-1} at 298.15 K) and was used as solvent for the preparation of 0.02, 0.04, 0.06, 0.08, and 0.10 m solutions of glucose, sucrose, and raffinose. The molalities of the solutions prepared were within an accuracy of ±0.0001 mol kg^{-1}. The solutions were stored in special air tight bottles to avoid the absorption of atmospheric moisture and carbon dioxide, if any. All the solutions were prepared afresh and weighing was done on a Precisa XB-220 (Swiss-make) electronic balance with a precision of $\pm1\times10^{-4}$ g.

The densities of the solutions were measured using a pycnometer (Borosil glass, total volume of 8×10^{-6} m^3) having a graduated capillary of narrow bore (internal diameter 1×10^{-3} m). The capillary was provided with a well-fitted glass cap in order to avoid changes in composition due to evaporation. The marks on the pycnometer were calibrated at experimental temperatures using known densities of double distilled deionised water and extra pure ethanol (E. Merck, Germany, highly pure). The accuracy of the density measurement was checked by comparing the experimental values of the densities of water and ethanol and good agreement was found with the corresponding literature values (Stokes and Mills, 1965; Ali and Nain, 1997). Details regarding calibration, experimental set up and procedure have been described elsewhere (Parfenyuk et al., 2004; Ali and Nain, 2002; Pal and Kumar, 2005; Behrends et al., 2002). Density measurements were made in triplicate and an average value was used for all the calculations. The uncertainty in density measurement was less than $\pm5\times10^{-2}$ kg m^{-3}. The viscosity measurements were made by using a thoroughly cleaned Ubbelohde type suspended-level viscometer with a flow time of 300 s for pure water at 298.15 K. Since the flow time was greater than 100 s, kinetic energy corrections were not considered. The time of flow was recorded with a digital stop-watch, accurate to ±0.1 s. An average of four sets of flow times for each reading was taken and used for the calculation of viscosity of the solutions. The accuracy in viscosity measurement was found to be $\pm3\times10^{-4}$ N s m^{-2}. Refractive indices of the solutions were measured with the help of a thermostatic Abbe – refractometer. Before use, the refractometer was calibrated with double distilled deionised water and toluene (E. Merck, India, mass fraction 0.99) at experimental temperatures. The accuracy in refractive index measurement was up to ±0.0002 units. The temperature of the solutions during the measurements of ρ, η, and n_D was maintained (±0.02 K) in an electronically controlled thermostatic water bath (JULABO, Germany).

3. Results and discussion

The density (ρ), viscosity (η), and refractive index (n_D) measured for the solutions of glucose, sucrose, and raffinose in aqueous glycine at 298.15, 303.15, 308.15, and 313.15 K are listed in Table 1.

3.1. Volumetric study

Apparent molar volumes, V_ϕ of glucose, sucrose, and raffinose in 0.015 m aqueous glycine at 298.15, 303.15, 308.15, and 313.15 K were calculated from the experimentally measured densities using the following relation (Millero et al., 1978; Ali et al., 2008):

$$V_\phi = M/\rho - 10^3(\rho-\rho_o)/m\rho\rho_o \quad (1)$$

where M and m are the molar mass of the solute and molality of the solution, respectively, ρ and ρ_o are the densities of the solution and solvent, respectively. The calculated values of V_ϕ of saccharides in water and in aqueous glycine solution as a function of concentration and temperature are presented in Table 2. Apparent molar volume reflects the size of the hydrated molecules in solution and thus the interaction of solute molecules with the solvent (Parke et al., 1999).

Table 1 Densities, ρ, viscosities, η, and refractive indices, n_D of glucose, sucrose, and raffinose in aqueous glycine at different temperatures.

m (mol kg^{-1})	T/(K)			
	298.15	303.15	308.15	313.15
ρ (kg m^{-3})				
Glucose + water				
0.01	997.77	996.36	994.74	992.92
0.02	998.44	997.03	995.41	993.58
0.03	999.12	997.70	996.08	994.25
0.04	999.79	998.37	996.74	994.91
0.05	1000.5	999.03	997.41	995.56
Sucrose + water				
0.01	998.38	996.98	995.36	993.54
0.02	999.68	998.27	996.64	994.81
0.03	1001.0	999.55	997.91	996.07
0.04	1002.3	1000.8	999.17	997.32
0.05	1003.5	1002.1	1000.4	998.56
Raffinose + water				
0.01	999.05	997.65	996.03	994.21
0.02	1001.0	999.60	997.98	996.15
0.03	1002.9	1001.5	999.91	998.07
0.04	1004.8	1003.4	1001.8	999.98
0.05	1006.7	1005.3	1003.7	1001.9
Glucose + aq. Glycine				
0.00	997.56	996.15	994.53	992.70
0.02	998.91	997.49	995.86	994.02
0.04	1000.3	998.82	997.18	995.32
0.06	1001.6	1000.1	998.49	996.63
0.08	1002.9	1001.5	999.79	997.92
0.10	1004.2	1002.7	1001.1	999.20
Sucrose + aq. Glycine				
0.00	997.56	996.15	994.53	992.70
0.02	1000.1	998.71	997.08	995.24
0.04	1002.7	1001.3	999.60	997.75
0.06	1005.1	1003.7	1002.1	1000.2
0.08	1007.4	1006.3	1004.5	1002.6
0.10	1009.8	1008.7	1006.9	1005.1
Raffinose + aq. Glycine				
0.00	997.56	996.15	994.53	992.70
0.02	1001.5	1000.0	998.36	996.53
0.04	1005.3	1003.9	1002.1	1000.2
0.06	1009.1	1007.6	1005.7	1003.9
0.08	1012.8	1011.2	1009.3	1007.6
0.10	1016.4	1014.8	1012.8	1011.0
η (10^{-3} N s m^{-2})				
Glucose + aq. Glycine				
0.00	0.8870	0.7983	0.7187	0.6528
0.02	0.9028	0.8080	0.7257	0.6574
0.04	0.9195	0.8178	0.7328	0.6621
0.06	0.9361	0.8278	0.7399	0.6668
0.08	0.9526	0.8377	0.7471	0.6715
0.10	0.9671	0.8472	0.7539	0.6759
Sucrose + aq. Glycine				
0.00	0.8870	0.7983	0.7187	0.6528
0.02	0.9035	0.8121	0.7275	0.6586
0.04	0.9196	0.8248	0.7366	0.6648
0.06	0.9372	0.8398	0.7456	0.6704
0.08	0.9555	0.8550	0.7548	0.6762
0.10	0.9701	0.8669	0.7636	0.6827
Raffinose + aq. Glycine				
0.00	0.8870	0.7983	0.7187	0.6528
0.02	0.9110	0.8196	0.7331	0.6663

(*continued on next page*)

Table 1 (*continued*)

m (mol kg^{-1})	T/(K)			
	298.15	303.15	308.15	313.15
0.04	0.9396	0.8421	0.7491	0.6783
0.06	0.9695	0.8647	0.7651	0.6914
0.08	1.0002	0.8874	0.7813	0.7085
0.10	1.0011	0.9098	0.7929	0.7198
n_D				
Glucose + aq. Glycine				
0.00	1.3322	1.3316	1.3314	1.3310
0.02	1.3325	1.3324	1.3318	1.3314
0.04	1.3327	1.3325	1.3320	1.3315
0.06	1.3335	1.3334	1.3329	1.3324
0.08	1.3337	1.3335	1.3331	1.3328
0.10	1.3345	1.3335	1.3330	1.3335
Sucrose + aq. Glycine				
0.00	1.3322	1.3316	1.3314	1.3310
0.02	1.3332	1.3324	1.3323	1.3318
0.04	1.3345	1.3344	1.3333	1.3327
0.06	1.3355	1.3345	1.3344	1.3340
0.08	1.3366	1.3359	1.3351	1.3346
0.10	1.3367	1.3366	1.3357	1.3355
Raffinose + aq. Glycine				
0.00	1.3322	1.3316	1.3314	1.3310
0.02	1.3365	1.3417	1.3355	1.3345
0.04	1.3376	1.3372	1.3368	1.3361
0.06	1.3392	1.3386	1.3382	1.3375
0.08	1.3405	1.3396	1.3394	1.3386
0.10	1.3417	1.3415	1.3409	1.3400

Apparent specific volumes, $V_{\phi,sp}$ for glucose, sucrose, and raffinose in water and in aqueous glycine at different temperatures have been calculated by the following equation (Parke and Birch, 1999; Mathlouthi et al., 1996).

$$V_{\phi,sp} = V_\phi / M \tag{2}$$

The values of $V_{\phi,sp}$ are included in Table 2. Apparent specific volumes measure the displacement of solvent by solute and reflect the compatibility of solute with solvent structure (Chavez and Birch, 1997). It is also a measure of taste quality, for sweet molecules, the range of apparent specific volume lies in the range 0.55–0.68 cm^3 g^{-1}, and for substances which exhibit a clean sweet taste, their values lies in the range 0.60–0.64 cm^3 g^{-1} (Parke et al., 1999). The value of $V_{\phi,sp}$ (Table 2) of the saccharides studied increases with an increase in concentration and temperature of the solution, and follows the order sucrose < glucose < raffinose in water as well as in aqueous glycine except at 298.15 K where the order is glucose < sucrose < raffinose. This suggests that glucose molecules are more hydrated than sucrose and raffinose in water as well as in glycine, as glucose has the lowest $V_{\phi,sp}$. The saccharide molecules are more hydrated in water than in aqueous glycine. The increase of $V_{\phi,sp}$ with the increase in concentration of the solute suggests that their hydration decreases, in water and in aqueous glycine and the probability of saccharide–saccharide and saccharide–glycine interaction increases respectively.

It can be seen from Table 2 that V_ϕ is found to vary linearly with concentration at all temperatures and thus the data are fitted to the Eq. (3) to determine the partial molar volume at infinite dilution, V_ϕ^0, by a regression analysis based on least squares method (Ali et al., 2008).

$$V_\phi = V_\phi^0 + S_v m \tag{3}$$

where S_v is the experimental slope which is also considered as the volumetric pair wise interaction coefficient (Ali and Nain, 2002; Ali et al., 2008; Ali and Shahjahan, 2008) and V_ϕ^0, the experimental intercept, is the limiting partial molar volume at infinite dilution of the saccharides in water and in aqueous glycine. Partial molar volume at infinite dilution, V_ϕ^0, reflects the strength of solute–solvent interactions, whereas S_v can serve as a quantitative estimate of solute–solute interactions (Ali and Nain, 2002; Ali et al., 2008). The respective data are presented in Table 3. The high positive V_ϕ^0 values for all the three saccharides in water as well as in aqueous glycine at each studied temperature indicate the presence of strong solute–solvent i.e., glucose/sucrose/raffinose–glycine–water interactions. The values of V_ϕ^0 increase in the following order: glucose < sucrose < raffinose in water as well as in aqueous glycine. As V_ϕ^0 also provides important information of solute hydrophobicity, hydration properties apart from solute–solvent interactions (Ali and Shahjahan, 2008), an increase in V_ϕ^0 from glucose to raffinose indicates the enhancement of sugar rings, hydroxyl groups as well as the hydrophobicity of saccharides. The V_ϕ^0 values for glucose/sucrose/raffinose in aqueous glycine are greater than the corresponding values in pure water (Table 3), suggesting that glucose/sucrose/raffinose interact more strongly with glycine/water molecules than they do in

Table 2 Apparent molar volumes, V_ϕ and apparent specific volumes, $V_{\phi,sp}$ of glucose, sucrose, and raffinose in aqueous glycine at different temperatures.

m (mol kg^{-1})	T/(K)			
	298.15	303.15	308.15	313.15
V_ϕ (10^{-6} m^3 mol^{-1})				
Glucose + water				
0.01	110.70	112.07	112.64	113.13
0.02	111.88	112.60	112.92	113.72
0.03	111.89	112.72	112.96	113.52
0.04	112.11	112.75	113.20	113.64
0.05	111.40	112.93	113.11	113.88
Sucrose + water				
0.01	211.76	212.17	212.81	213.36
0.02	211.73	212.50	213.39	214.26
0.03	210.87	212.77	213.74	214.71
0.04	210.80	213.52	214.04	215.06
0.05	212.68	212.85	214.71	215.36
Raffinose + water				
0.01	396.81	397.39	398.22	398.99
0.02	397.29	397.72	398.29	399.37
0.03	398.62	399.00	398.48	399.66
0.04	398.90	399.26	399.21	399.68
0.05	398.77	399.12	399.14	399.18
Glucose + aq. Glycine				
0.02	112.61	113.18	113.76	114.35
0.04	111.45	113.28	113.86	114.71
0.06	112.48	114.06	113.96	114.56
0.08	112.92	112.85	114.07	114.66
0.10	113.12	114.09	113.97	114.77
Sucrose + aq. Glycine				
0.02	214.96	214.08	214.72	215.39
0.04	212.91	212.77	214.94	215.60
0.06	215.22	215.18	214.98	216.33
0.08	217.39	213.59	216.01	217.07
0.10	217.47	214.45	216.42	216.28
Raffinose + aq. Glycine				
0.02	398.45	401.77	402.62	403.00
0.04	398.43	398.46	403.37	405.55
0.06	398.08	399.90	405.01	404.89
0.08	398.44	401.16	405.10	403.82
0.10	399.10	401.35	405.61	405.70
$V_{\phi,sp}$ (10^{-3} m^3 kg^{-1})				
Glucose + water				
0.01	0.614	0.622	0.625	0.628
0.02	0.621	0.625	0.627	0.631
0.03	0.621	0.626	0.627	0.630
0.04	0.622	0.626	0.628	0.631
0.05	0.618	0.627	0.628	0.632
Sucrose + water				
0.01	0.619	0.620	0.622	0.623
0.02	0.619	0.621	0.623	0.626
0.03	0.616	0.622	0.624	0.627
0.04	0.616	0.624	0.625	0.628
0.05	0.621	0.622	0.627	0.629
Raffinose + water				
0.01	0.667	0.668	0.670	0.671
0.02	0.668	0.669	0.670	0.672
0.03	0.670	0.671	0.670	0.672
0.04	0.671	0.672	0.671	0.672
0.05	0.671	0.671	0.671	0.671

(*continued on next page*)

Table 2 (*continued*)

m (mol kg^{-1})	T/(K)			
	298.15	303.15	308.15	313.15
Glucose + aq. Glycine				
0.02	0.625	0.628	0.631	0.635
0.04	0.619	0.629	0.632	0.637
0.06	0.624	0.633	0.633	0.636
0.08	0.627	0.626	0.633	0.636
0.10	0.628	0.633	0.633	0.637
Sucrose + aq. Glycine				
0.02	0.628	0.625	0.627	0.629
0.04	0.622	0.622	0.628	0.630
0.06	0.629	0.629	0.628	0.632
0.08	0.635	0.624	0.631	0.634
0.10	0.635	0.626	0.632	0.632
Raffinose + aq. Glycine				
0.02	0.670	0.676	0.677	0.678
0.04	0.670	0.670	0.678	0.682
0.06	0.670	0.673	0.681	0.681
0.08	0.670	0.675	0.681	0.679
0.10	0.671	0.675	0.682	0.682

Table 3 Limiting partial molar volume, V_ϕ^0, slope, S_v, limiting partial molar volume in water, $V_{\phi,water}^0$, volume of transfer, $\Delta_{tr}V_\phi^0$ of glucose, sucrose, and raffinose in aqueous glycine at different temperatures.

Parameter	T/(K)			
	298.15	303.15	308.15	313.15
Glucose + aq. Glycine				
V_ϕ^0 (10^{-6} m^3 mol^{-1})	111.775	113.074	113.739	114.377
S_v (10^{-6} m^3 mol^{-2} kg)	12.35	6.99	3.11	3.92
$V_{\phi,water}^0$ (10^{-6} m^3 mol^{-1})	111.11	112.05	112.61	113.15
	112.7[a]	113.3[a]	113.5[a]	113.9[a]
	111.89[b]	–	112.81[b,d]	–
	111.87[d]	–	112.7[c]	–
	112.91[e]	–	112.82[e]	–
$\Delta_{tr}V_\phi^0$ (10^{-6} m^3 mol^{-1})	0.67	1.03	1.13	1.23
Sucrose + aq. Glycine				
V_ϕ^0 (10^{-6} m^3 mol^{-1})	212.75	213.55	214.07	215.16
S_v (10^{-6} m^3 mol^{-2} kg)	47.41	7.74	22.40	16.28
$V_{\phi,water}^0$ (10^{-6} m^3 mol^{-1})	211.29	212.05	212.40	213.11
	211.87[b]	–	212.75[b]	–
	211.91[d]	–	213.5[c]	–
	211.92[e]	–	212.70[d]	–
	–	–	212.74[e]	–
$\Delta_{tr}V_\phi^0$ (10^{-6} m^3 mol^{-1})	1.45	1.50	1.67	2.05
Raffinose + aq. Glycine				
V_ϕ^0 (10^{-6} m^3 mol^{-1})	398.10	399.97	402.03	403.49
S_v 10^{-6} m^3 mol^{-2} kg)	6.62	9.31	38.61	18.37
$V_{\phi,water}^0$ (10^{-6} m^3 mol^{-1})	396.41	397.00	397.84	399.17
	397.11[b]	–	398.93[b]	–
	397.09[d]	–	398.96[d]	–
	397.10[e]	–	398.97[e]	–
$\Delta_{tr}V_\phi^0$ (10^{-6} m^3 mol^{-1})	1.69	2.97	4.19	4.32

[a] Banipal et al. (2010).
[b] Dhondge et al. (2011).
[c] Banipal et al. (2009).
[d] Banipal et al. (1997).
[e] Zhuo et al. (2006).

aqueous solution. It is also found that V_ϕ^0 values increase with an increase in temperature due to reduced electrostriction, i.e., weakening of saccharide–water hydrogen bonds which tend to release water molecules from the hydration layers of the solute to the bulk, resulting in increased V_ϕ^0 values with a rise in temperature (Ali et al., 2008). The dominance of solute–solvent interactions has also been observed (Roy et al., 2010) for saccharides in aqueous cetyltrimethylammonium bromide. In our studied systems S_v values (Table 3) are much smaller than V_ϕ^0 values which imply weak solute–solute interactions.

The partial molar volumes of transfer of saccharides from water to aqueous glycine at infinite dilution $\Delta V_{\phi(tr)}^0$ have been calculated from V_ϕ^0 values by the equation:

$$\Delta V_{\phi(tr)}^0 = V_\phi^0(aq.glycine) - V_\phi^0(water) \quad (4)$$

The values of partial molar volumes of saccharides in water $V_\phi^0(water)$ are collected from the literature values, (Paljk et al., 1990; Banipal et al., 2010; Banipal et al., 2009, 1997; Dhondge et al., 2011; Zhuo et al., 2006) are also given in Table 3. The calculated values of $\Delta V_{\phi(tr)}^0$ at all studied temperatures are summarised in Table 3 which indicates that V_ϕ^0 of the studied saccharides in aqueous glycine are greater than those in pure water, i.e., $\Delta V_{\phi(tr)}^0$ values are positive for all the three investigated saccharides. Positive transfer volumes for saccharides have also been observed by other workers (Banipal et al., 2010; Banipal et al., 2008; Banipal et al., 2002; Banipal et al., 2000; Zhuo et al., 2006). Again, like V_ϕ^0, the values of $\Delta V_{\phi(tr)}^0$ for the studied saccharides follow the order glucose < sucrose < raffinose, which means the transfer volumes increase with an increase in molecular complexity from mono- to tri-saccharide. The view presented here regarding close relationship between molecular structure of saccharides and their transfer volumes is in good agreement with the findings reported in the literature (Dhondge et al., 2011; Banipal et al., 1997) that $\Delta V_{\phi(tr)}^0$ tends to increase with an increase in the complexity of the molecules. Furthermore, the

transfer volumes can also be explained by the co-sphere overlap model (Freidman and Krishnan, 1973). The following types of possible interactions are expected between saccharides and glycine molecules:

1. Hydrophilic-ionic interactions between the (–OH, –C═O, and –O–) groups of saccharides and the zwitterionic centres (NH_3^+, COO) of glycine.
2. Hydrophilic–hydrophobic interactions between the OH groups of saccharides and the nonpolar group of glycine.
3. Hydrophobic–hydrophobic interactions between the nonpolar groups of saccharides and nonpolar group of glycine.

The interactions of type (1) make positive contribution to the transfer volume, whereas, the contributions of types (2) and (3) lead to a negative contribution to $\Delta V^0_{\phi(tr)}$. Therefore, the observed increase in positive transfer volumes suggests that hydrophilic-ionic interactions are predominant in the ternary systems studied. It is well known that (Umemura et al., 2005) hydrophilic hydroxyl groups of saccharides induce a considerable contraction in volume of the peripheral water molecules due to hydrogen bond network formation, which is subsequently diminished on the addition of glycine because of the saccharide–glycine interactions involving (NH_3^+, COO^-) groups of glycine and (–OH, –C═O, and –O–) groups of glucose, sucrose, and raffinose, releasing water molecules from their bond states to the bulk of solution and, thus, making $\Delta V^0_{\phi(tr)}$ positive. Similar conclusions were also drawn by others for saccharides in aqueous salts (Banipal et al., 2010; Banipal et al., 2008; Banipal et al., 2002; Banipal et al., 2000; Zhuo et al., 2006) and in aqueous surfactant (Roy et al., 2010) solutions.

The variation of V^0_ϕ values with temperature can be expressed through the following relation:

$$V^o_\phi = a + bT + cT^2 \tag{5}$$

where a, b, and c are constants and T is the temperature in Kelvin. Values of these constants along with the regression coefficients are given in Table 4. The limiting partial molar expansibility E^0_ϕ can be obtained by the following relation.

$$E^0_\phi = \left(\partial V^0_\phi/\partial T\right)_P = b + 2cT \tag{6}$$

The values of E^0_ϕ are listed in Table 5. A close inspection of the Table 5 shows that the E^0_ϕ values are positive for all the studied systems, decrease for glucose and raffinose in aqueous glycine, while, exhibit an opposite trend for sucrose with a rise in temperature. Decrease in E^0_ϕ for glucose and raffinose with temperature is ascribed to the release of solvent molecules from the hydration spheres of glucose and raffinose due to the increased thermal energy of the molecules. This would lead to the increased solute–solute interactions. An opposite behaviour is observed for sucrose in aqueous glycine. Similar behaviour has been reported for amino acids in water + dimethyl sulphoxide solutions (Dash and Pasupalak, 1997). Following Hepler's (Hepler, 1969) method of examining the structure–making or – breaking capacity of a solute on the basis of the sign of $(\partial^2 V^0_\phi/\partial T^2)_P$. It can be shown using general thermodynamics that

$$\left(\partial C^0_p/\partial P\right)_T = -T\left(\partial^2 V^0_\phi/\partial T^2\right)_P \tag{7}$$

where C^0_p is the partial molar heat capacity at infinite dilution. A structure-making solute should have positive values of $(\partial^2 V^0_\phi/\partial T^2)_P$ while negative values of $(\partial^2 V^0_\phi/\partial T^2)_P$ are shown by structure-breaking solutes. In the light of this reasoning, we observe that glucose and sucrose having positive $(\partial^2 V^0_\phi/\partial T^2)_P$ values (Table 5) act as structure-makers, whereas, raffinose with a negative value behaves as a structure-breaker in aqueous solutions. In the presence of glycine the values of $(\partial^2 V^0_\phi/\partial T^2)_P$ are positive for glucose and raffinose while it is negative in the case of sucrose, indicating that the first two saccharides behave as structure-makers whereas the third one acts as a structure-breaker.

3.2. Viscometric study

The data on viscosities, η (Table 1) are also graphically displayed in Fig. 1. It reveals that η tends to increase linearly with an increase in the concentration of saccharides at each investigated temperature. This clearly indicates increased saccharide – glycine/water interaction as the amount of saccharide increases in the solution. Moreover, at a given temperature, the value of η increases from glucose to raffinose in the sequence: glucose < sucrose < raffinose, which, in turn, suggests the increased strength of saccharide – glycine/water interaction. As the temperature of the system increases the value of η decreases for each studied saccharide. Increase in thermal energy of the system, due to rise in temperature, promotes the breaking up of saccharide–glycine/water aggregates. This facilitates the flow of the system, making η decrease with temperature.

The viscosity data (Table 1) for saccharides in aqueous glycine at different temperatures have been analysed by using the expression (Jones and Dole, 1929):

$$\eta_r = \eta/\eta_0 = 1 + Ac^{1/2} + Bc \tag{8}$$

where η_r is the relative viscosity, η_0 and η are the viscosities of solvent (aqueous glycine) and solution, respectively, and c is the molarity calculated from molality data (Motin, 2004) for saccharides. A, Falkenhagen coefficient reflects the solute–solute interactions, whereas, B, the Jones–Dole coefficient,

Table 4 The values of algebraic coefficients for Eq. (5).

	a (10^{-6} m^3 mol^{-1})	b (10^{-6} m^3 mol^{-1} K^{-1})	c (10^{-6} m^3 mol^{-1} K^{-2})	r
Glucose + water	−302.1675	2.5788	−0.0040	0.99858
Sucrose + water	130.0006	0.4219	−0.0005	0.99134
Raffinose + water	1032.9454	−4.3412	0.0074	0.99966
Glucose + aq. Glycine	−555.0989	4.2046	−0.0066	0.99759
Sucrose + aq. Glycine	437.3397	−1.6178	0.0029	0.99408
Raffinose + aq. Glycine	−93.4442	2.8709	−0.0041	0.99906

Table 5 Values of E_ϕ^0 and $(\partial C_p^0/\partial P)_T$ for glucose, sucrose, and raffinose in aqueous glycine at different temperatures.

Parameter	T/(K)			
	298.15	303.15	308.15	313.15
Glucose + water				
E_ϕ^0 (10^{-6} m^3 mol^{-1} K^{-1})	0.19	0.15	0.11	0.07
$(\partial C_p^0/\partial P)_T$ (10^{-6} m^3 mol^{-1} K^{-1})	2.39	2.43	2.47	2.51
Sucrose + water				
E_ϕ^0 (10^{-6} m^3 mol^{-1} K^{-1})	0.12	0.12	0.11	0.11
$(\partial C_p^0/\partial P)_T$ (10^{-6} m^3 mol^{-1} K^{-1})	0.30	0.30	0.31	0.31
Raffinose + water				
E_ϕ^0 (10^{-6} m^3 mol^{-1} K^{-1})	0.07	0.15	0.22	0.29
$(\partial C_p^0/\partial P)_T$ (10^{-6} m^3 mol^{-1} K^{-1})	−4.41	−4.49	−4.56	−4.63
Glucose + aq. Glycine				
E_ϕ^0 (10^{-6} m^3 mol^{-1} K^{-1})	0.27	0.20	0.14	0.07
$(\partial C_p^0/\partial P)_T$ (10^{-6} m^3 mol^{-1} K^{-1})	3.94	4.00	4.07	4.13
Sucrose + aq. Glycine				
E_ϕ^0 (10^{-6} m^3 mol^{-1} K^{-1})	0.11	0.14	0.17	0.20
$(\partial C_p^0/\partial P)_T$ (10^{-6} m^3 mol^{-1} K^{-1})	−1.73	−1.76	−1.79	−1.82
Raffinose + aq. Glycine				
E_ϕ^0 (10^{-6} m^3 mol^{-1} K^{-1})	0.43	0.39	0.34	0.30
$(\partial C_p^0/\partial P)_T$ (10^{-6} m^3 mol^{-1} K^{-1})	2.44	2.49	2.53	2.57

provides information regarding solute–solvent interactions in the solution. *A*- and *B*-coefficients were obtained by the least-squares method as intercept and slope of the linear plot of $(\eta_r - 1/c^{1/2})$ against $c^{1/2}$ and are summarised in Table 6.

An inspection of Table 6 shows that *B*-coefficients are larger than *A*-coefficients, suggesting the presence of strong solute–solvent interactions as compared to weak solute–solute interactions, hence supporting the behaviours of V_ϕ^0 and S_v, respectively. In our studied systems, the observed values of *B*-coefficients are positive for all the saccharides at each investigated temperature, follow the sequence: glucose < sucrose < raffinose. This suggests increased saccharide–glycine/water interaction and that the strength of the interaction follows the above sequence. This is explained by considering that as the number of hydroxyl groups increases from glucose to raffinose so does the number of H-bonds formed between saccharides and water/glycine molecules, (Mishra et al., 1997) resulting in increased *B*-values in the sequence mentioned above.

The hydration number, n_H of saccharides studied is obtained by the relation (Mishra et al., 1997; Linow and Philipp, 1984):

$$n_H = \left(\overline{V_\eta} - \overline{V_2^0}\right)/\overline{V_1^0} \tag{9}$$

where $\overline{V_\eta}$ is the volume of the hydrated solute and can be obtained as the slope of the plot of η_{sp}/c vs. η_{sp} (specific viscosity of the solution) (Linow and Philipp, 1984). $\overline{V_2^0}$ and $\overline{V_1^0}$ are the partial molar volumes of solute and solvent, respectively. Hydration numbers vary with the method used to calculate them, (Parke and Birch, 1999) so it is not fair enough to compare the values obtained by different methods. Thus, it is reported that for saccharides n_H may range from 1.8 (NMR) to 21 (NIR), (Allen et al., 1974) for sucrose in aqueous solutions, its values are reported as 5.3 (intrinsic viscosity), (Bressan and Mathlouthi, 1994) 5 (activity measurement), (Akhumov, 1981) and 13.8 (ultrasonic velocity data) (Chen et al., 1981). The value of hydration numbers of the saccharides investigated is given in Table 6. The hydration number mainly comes from the hydrogen bond formation by hydroxyl groups of saccharides with water as these hydroxyl groups have strong hydration ability. The hydration number so obtained follows the order glucose < sucrose < raffinose, but the computed values (Table 6) are not showing a proportional increase in the hydration number with the number of hydroxyl groups in glucose, sucrose, and raffinose which lead us to conclude that all the hydroxyl groups are not freely available for hydration with water. The compatibility of the studied saccharides with the 3-D hydrogen bond structure of water depends not only on the number of hydroxyl groups and oxygen atoms but also on the position (axial or equatorial) of the hydroxyl groups. In our system glucose is a monosaccharide with three equatorial and one axial hydroxyl group (Chen et al., 1981). At 298.15 K the n_H for glucose is approximately 9 which is quite a large number suggesting that the hydroxyl groups of glucose are available for hydrogen bonding with water molecules and hence pack well within the structure of water. The sucrose molecule consists of glucose and fructose monosaccharide units and contains two intramolecular hydrogen bonds (Seuvre and Mathlouthi, 2010). It has a large number of equatorial hydroxyl groups, which might be the reason for its higher hydration (approximately 11 at 298.15 K) than glucose, whereas raffinose is a trisaccharide in which galactose is connected to the glucosyl group of sucrose via α-1, 6 linkage (Cheng and Lin, 2006). It's n_H at 298.15 K is approximately 15, which is larger than the n_H of glucose and sucrose. This suggests that though the value of n_H increases with the increase in the number of –OH groups as we move from glucose to raffinose, the increase is not in proportion to the number of hydroxyl groups in these saccharides. This is due to the fact

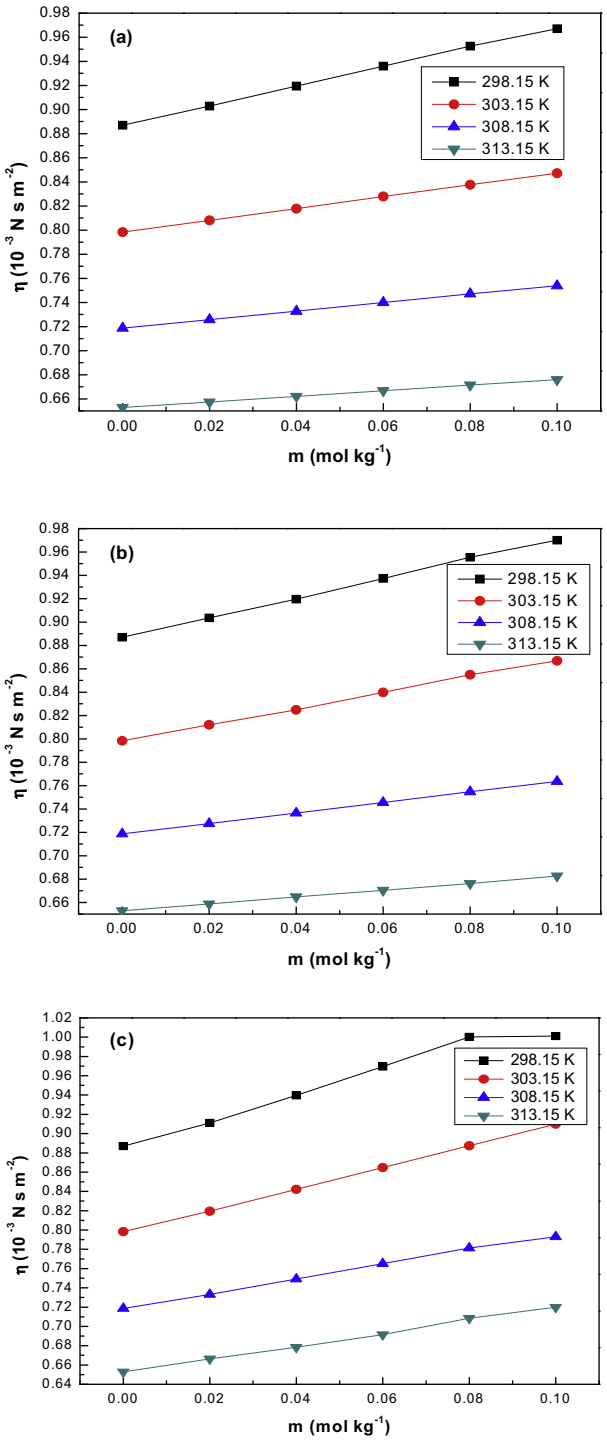

Figure 1 Plots of viscosities, η versus molality, m of (a) glucose, (b) sucrose, and (c) raffinose in aqueous glycine at different temperatures.

that steric hindrance is least for glucose and maximum for raffinose molecules for the formation of H-bonds with water molecules.

The free energy of activation of viscous flow per mole of solvent $\Delta\mu_1^{0\#}$ was calculated using the relation proposed by Eyring and co-workers (Glasstone et al., 1941):

Table 6 Values of A- and B-coefficients, n_H, $\Delta\mu_1^{0\#}$, and $\Delta\mu_2^{0\#}$ of glucose, sucrose, and raffinose in aqueous glycine at different temperatures.

Parameter	T/(K)			
	298.15	303.15	308.15	313.15
Glucose + aq. Glycine				
A (10^{-2} dm$^{3/2}$ mol$^{-1/2}$)	−0.41	−0.27	−0.17	−0.10
B (dm^3 mol^{-1})	0.93	0.63	0.50	0.36
n_H	8.65	5.27	3.01	1.84
$\Delta\mu_1^{0\#}$ (kJ mol^{-1})	60.52	61.27	62.02	62.78
$\Delta\mu_2^{0\#}$ (kJ mol^{-1})	201.79	162.00	146.33	128.45
Sucrose + aq. Glycine				
A (10^{-2} dm$^{3/2}$ mol$^{-1/2}$)	−1.41	−1.26	−0.80	−0.53
B (dm^3 mol^{-1})	1.02	0.94	0.67	0.49
n_H	10.64	8.93	5.62	2.46
$\Delta\mu_1^{0\#}$ (kJ mol^{-1})	60.52	61.27	62.02	62.78
$\Delta\mu_2^{0\#}$ (kJ mol^{-1})	227.72	219.14	184.88	160.70
Raffinose + aq. Glycine				
A (10^{-2} dm$^{3/2}$ mol$^{-1/2}$)	0.14	−2.76	−1.65	−1.90
B (dm^3 mol^{-1})	1.48	1.53	1.15	1.12
n_H	14.9	11.17	8.96	5.12
$\Delta\mu_1^{0\#}$ (kJ mol^{-1})	60.52	61.27	62.02	62.78
$\Delta\mu_2^{0\#}$ (kJ mol^{-1})	316.18	327.78	278.59	278.84

$$\eta_0 = (hN_A/\overline{V}_1^0)\ \exp(\Delta\mu_1^{0\#}/RT) \tag{10}$$

where h, N_A, and $\overline{V}_1^0$ are the Planck's constant, Avogadro number, and partial molar volume of the solvent, respectively, and R, the universal gas constant. The rearrangement of Eq. (10) yields:

$$\Delta\mu_1^{0\#} = RT\ln\left(\eta_0\overline{V}_1^0/hN_A\right) \tag{11}$$

The free energy of activation of viscous flow per mole of solute $\Delta\mu_2^{0\#}$ can be calculated by using the (Feakins et al., 1974, 1993) extension of Eyring transition-state theory:

$$B = \left(\overline{V}_1^0 - \overline{V}_2^0\right)/1000 + \overline{V}_1^0\left[\left(\Delta\mu_2^{0\#} - \Delta\mu_2^{0\#}\right)/1000RT\right] \tag{12}$$

where $\overline{V}_2^0(=V_\phi^0)$ is the partial molar volume of the solute. The rearrangement of the above equation gives:

$$\Delta\mu_2^{0\#} = \Delta\mu_1^{0\#} + \left(RT/\overline{V}_1^0\right)\left[1000B - \left(\overline{V}_1^0 - \overline{V}_2^0\right)\right] \tag{13}$$

The computed values of $\Delta\mu_1^{0\#}$ and $\Delta\mu_2^{0\#}$ at different temperatures are given in Table 6. It reveals that the values of $\Delta\mu_2^{0\#}$ are much larger than $\Delta\mu_1^{0\#}$ values which suggest that the formation of the transition state is less favoured in aqueous glycine and that there are strong glucose/sucrose/raffinose–solvent (glycine + water) interactions in the ground state than in the transition state, which might be because of the breaking and distortion of intermolecular bonds in the latter state. The values of $\Delta\mu_2^{0\#}$ of studied saccharides are increasing in the order glucose < sucrose < raffinose. This implies that more energy is required for the saccharides having more sugar units with complex structure in transferring from ground state to the transition state.

The free energy of activation of viscous flow of solution $\Delta G^{0\#}$ was evaluated from the relation:

$$\Delta G^{0\#} = n_1\Delta\mu_1^{0\#} + n_2\Delta\mu_2^{0\#} \tag{14}$$

Table 7 Enthalpies, $\Delta H^{0\#}$ and entropies, $\Delta S^{0\#}$ of activation of viscous flow of solutions of glucose, sucrose, and raffinose in aqueous glycine at different temperatures.

m (mol kg^{-1})	$\Delta H^{0\#}$ (kJ mol^{-1})	$\Delta S^{0\#}$ (10^{-2} kJ mol^{-1} K^{-1})
Glucose + aq. Glycine		
0	0.23	0.23
0.02	9.35	−1.55
0.04	18.46	−3.32
0.06	27.57	−5.10
0.08	36.69	−6.87
0.10	45.80	−8.64
Sucrose + aq. Glycine		
0	0.23	0.23
0.02	32.97	−9.19
0.04	65.70	−18.60
0.06	98.43	−28.01
0.08	131.16	−37.43
0.10	163.90	−46.84
Raffinose + aq. Glycine		
0	0.23	0.23
0.02	25.95	−6.22
0.04	51.67	−12.67
0.06	77.39	−19.12
0.08	103.10	−25.57
0.10	128.82	−32.02

where n_1 and n_2 are the respective number of moles of binary solvent (aqueous glycine) and solute (glucose/sucrose/raffinose), respectively. The enthalpy, $\Delta H^{0\#}$ and entropy, $\Delta S^{0\#}$ of activation of viscous flow were calculated by using the equation:

$$\Delta G^{0\#} = \Delta H^{0\#} - T\Delta S^{0\#} \tag{15}$$

The values of $\Delta H^{0\#}$ and $\Delta S^{0\#}$ were obtained as intercept and slope, respectively, from the plots of $\Delta G^{0\#}$ vs T. The results are shown in Table 7. $\Delta H^{0\#}$ and $\Delta S^{0\#}$ are useful in providing the structural information about solute species and solute–solvent interactions. It is clear from the Table 7 that $\Delta H^{0\#}$ values are positive and increase with an increase in the concentration of glucose, sucrose and raffinose which indicates that the formation of activated species necessary for viscous flow seems to be difficult as the amount of these saccharides increases in aqueous glycine. The $\Delta S^{0\#}$ values are negative and exhibit a pronounced decrease with the increase in concentration of all the three saccharides, pointing out that the systems become more structured as a result of strong interactions between solute and solvent molecules.

3.3. Refractivity study

The molar refractivity, R_D of the mixtures under study can be calculated from the refractive indices n_D using Lorentz–Lorenz equation (Lorentz, 1880; Lorenz, 1880; Marcus, 1977; Ali et al., 2006):

$$R_D = \left[(n_D^2 - 1)/(n_D^2 + 1)\right]\left(\sum_{i=1}^{3} x_i M_i/\rho\right) \tag{16}$$

where x_i and M_i are the mole fraction and molecular weight of the i^{th} component of the mixture. The values of R_D are given in Table 8. It indicates that the values of all the three saccharides slightly increase with an increase in the concentration of saccharides. The trend in n_D and R_D values of the saccharides in aqueous glycine is: glucose < sucrose < raffinose. The refractive index of a substance indicates the packing of molecules in the solvent mixture (Banik and Roy, 2012). In the studied systems, we find that the refractive index and molar

Table 8 Values of molar refractive index, R_D for glucose, sucrose, and raffinose in aqueous glycine at different temperatures.

m (mol kg^{-1})	T/(K)			
	298.15	303.15	308.15	313.15
R_D (10^{-6} m^3 mol^{-1})				
Glucose + aq. Glycine				
0.00	5.04	5.04	5.05	5.05
0.02	5.06	5.06	5.06	5.07
0.04	5.07	5.07	5.07	5.08
0.06	5.09	5.09	5.09	5.10
0.08	5.10	5.10	5.11	5.11
0.10	5.12	5.11	5.12	5.13
Sucrose + aq. Glycine				
0.00	5.04	5.04	5.05	5.05
0.02	5.07	5.07	5.08	5.08
0.04	5.11	5.12	5.11	5.11
0.06	5.14	5.14	5.14	5.15
0.08	5.18	5.18	5.17	5.18
0.10	5.20	5.20	5.20	5.21
Raffinose + aq. Glycine				
0.00	5.04	5.04	5.05	5.05
0.02	5.13	5.21	5.14	5.13
0.04	5.19	5.19	5.19	5.19
0.06	5.25	5.25	5.25	5.25
0.08	5.30	5.30	5.31	5.30
0.10	5.36	5.36	5.37	5.36

refraction values are greater for raffinose compared to glucose and sucrose, indicating that the molecules are more tightly packed in the mixture resulting in maximum solute–solvent interactions in the case of raffinose followed by sucrose and least by glucose. This again supports the findings based on other studied parameters of glucose/sucrose/raffinose in aqueous glycine solutions.

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